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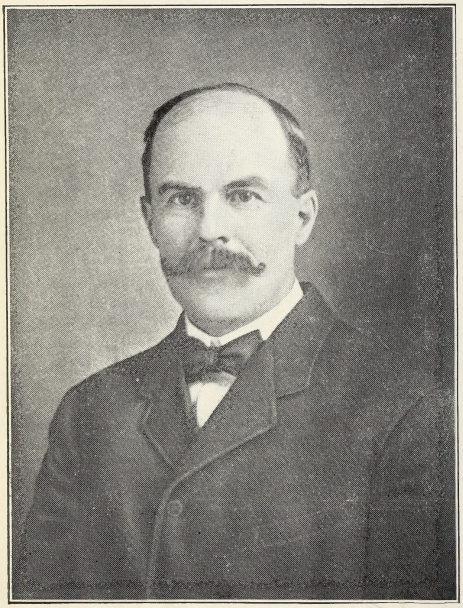


Prof. CHAS. B. WILSON

Biologist

Until 1932 Head of the Science Department at State Teachers College, Westfield, Professor Wilson, who was immensely popular with the students and among his confreres of the faculty, is a Biologist whose favourite forte is the study of marine and fresh-water life. He was a member of the Johns Hopkins Biological Expedition to Jamaica in 1897, and in '98-'99 was a research worker in the Tufts College Biological Laboratory at South Harpswell, Maine. From 1900 to 1923 Dr. Wilson served during summer vacations as temporary assistant of the U.S. Bureau of Fisheries. In this latter work the summers of 1900 to 1906 were spent at Woods Hole, Mass., at Beaufort, N.C., and at Lake Maxinkuckee, Ind.

In 1910 he went with another Johns Hopkins Expedition to Jamaica, and four months were devoted to the study of the parasites of tropical fishes. During 1911 a survey of the ponds and lakes of Minnesota, and during 1912 of the Cumberland River, was made in the interests of the pearl button manufacturing industry. The summers from 1913 to 1923 were spent at the Bureau of Fisheries at Fairport, Iowa, investigating the relation of water insects to fishpond culture, and subsequent years to similarly interesting research, including the identification, at Woods Hole, Mass, of the copepod plankton obtained during the last cruise of the ship "Carnegie" which cruised under the auspices of the Carnegie Institute of Washington, D.C. Since his retirement Professor Wilson intends to devote most of the year instead of summer only, to these researches into marine life.

Dr. Charles Branch Wilson was born in Exeter, Maine, Oct. 20, 1861, and received his education at Colby College, graduating in 1881 with the B.A. degree. Remaining at Colby as postgraduate student and tutor in Botany from 1881 to 1884 he received the M.A. in the latter year. In 1908 he was accorded the honorary degree of Sc.D.

Dr. Wilson became Professor of Natural Sciences at the State Normal School, Gorham, Maine, but resigned to make further studies at Johns Hopkins University, where he remained as student assistant 1895-6, and from which he received the degree of Ph.D. in 1910. In September, 1896, he was appointed Professor of Biology at the State Teachers College, Westfield, and in 1897 was made head of the Science Department, which included Geography. From this position he retired after 36 years of continuous work with but a single leave of absence.

EMBRYOLOGY.[1]

The Wrinkling of Frog's Eggs During Segmentation.—The occurrence of wrinkles in frog's eggs during the process of segmentation was first observed and very briefly described by Prevost and Dumas, who have the honor of being the first observers of the segmentation itself (Annales des Sciences Naturelles, I ser., 1824, Tom II, p. 110).

A somewhat better description was given later by Bär (Archiv. für Anatomie, etc., 1837) and Reichert (the same, 1841), who gave to the phenomenon the name "Faltenkranz," and made some attempt to explain its nature and origin.

By far the best description, and, indeed, the only really good one which has ever been published, is that by M. Schultze, which appeared in 1863 (Observationes nonnullæ de ovorum ranarum segmentatione, Bonnæ).

He gives an excellent account of wrinkles observed in the eggs of *Rana temporaria* and *R. esculenta*, and concludes with an "explanation of their origin." But he really devotes only a very few lines to the explanation, and gives up the remainder of this portion of his paper to

[1] Edited by E. A. Andrews, Baltimore, Md., to whom abstracts reviews and preliminary notes may be sent.

a controversy with Reichert, over the existence of a vitelline membrane on the surface of the egg.

Similar wrinkles have since been observed in the eggs of the common toad by Goette (Die Entwickelungsgeschichte der Unke), and of *Amblystoma punctatum* by Eycleshymer (Journal of Morphology, Vol X). But neither of these authors offer any explanation of their origin. So far as known this "Faltenkranz" has never been described for any American species of frog, nor has any attempt been made to study it by means of serial sections.

The following paper is offered as a contribution of some new and interesting details in the occurrence of the phenomenon, together with the results of a microscopic study of sections, in the hope of arriving at a rational conclusion as to its origin.

The author desires to acknowledge his great indebtedness to Dr. E. A. Andrews, for the suggestion which led to the study, and for much subsequent assistance. Thanks are also due to Prof. T. H. Morgan, for the kindly loan of a copy of Schultze's paper.

FORMATION OF THE WRINKLES.

The eggs of a small wood-frog, in all probability *Chorophilus triseriatus*, were obtained for class work on March 27, 1896.

They were unsegmented when found, and were immediately placed in ice water to check any further development.

After remaining thus for five hours they were used in the laboratory, being removed to watch-glasses containing tepid water. Some were allowed to remain in the ice water for eight hours before being examined, and it was noticed that these segmented much more rapidly than the ones which had been kept only five hours. Actual segmentation had been prevented during the stay in the ice water, but there seemed to have been a storing-up of energy, a sort of preparation for segmentation, so that when removed to a favorable environment the process began very quickly (5–10 mins.) and was carried on much faster than it would have been normally.

These eggs were obtained from pools covered with ice quarter of an inch thick, and most of the bunches were quite near the surface.

This must occur frequently where the eggs are laid so early in the spring; and according to Morgan (American Naturalist, August, 1891), Chorophilus always lays its eggs very early.

The storage of energy noticed above may suggest a natural method of compensation whereby the warm sunshine of mid-day may offset the freezing cold of the nights, and in this way the eggs will really lose very little time in their development.

O. Hertwig and Schultze have recently experimented on the influence of a very low temperature upon the development of the eggs of *Rana fusca* with very different results.

Hertwig (Sitzungsberichte der König, Preuss. Akad. d. Wiss, 5 April, 1894, p. 313) found that freshly fertilized eggs were injured by an exposure to a temperature of 0° C. for 24 hours. On being raised to the ordinary temperature a portion were developed very much more slowly than normal eggs, while in the remainder a part of the yolk was found incapable of division.

Schultze (Anatomischer Anzeiger, X Band, No. 9) subjected eggs of the same species to a temperature of 0° C. for 14 days, and then obtained perfectly normal embryos from them. These eggs had reached later stages of development before being cooled, and he does not state whether their subsequent development was more or less rapid than ordinary.

Subjection to a temperature of 0° C. for so long a period would probably have a very different effect from that of only a few hours duration.

Loeb and Norman (Archiv. f. Entwick, III Band, No. 1), experimenting on the eggs of the sea-urchin, Arbacia, found that when put in sea water to which had been added 2–3 per cent. NaCl or $MgCl_2$ segmentation of the protoplasm was wholly prevented, but that the nucleus went on dividing. On being put into normal sea water after a few hours exposure to the concentrated solution the eggs divided at once into several cells, the protoplasm merely rearranging itself around the new nuclear centers.

Possibly the same may be true in these frog's eggs when subjected for a short time to a freezing temperature, and the subsequent hastening of segmentation may be due to the fact that the nucleus has already divided.

In watching the segmentation of these eggs the great (comparative) size and depth of the furrows was specially noticed, together with the distinctness of the wrinkles formed along either side of them.

Accordingly it was determined to study their segmentation more in detail, and a fresh lot was obtained the next morning. These remained in ice water only one hour, just long enough to get them home. They were then transferred to watch-glasses and examined in strong sunlight. This fact must be taken into account in connection with the time periods given.

The first furrow appears at the superior pole without any previous flattening as in Amblystoma (Eycleshymer, Jour. of Morph., X, p. 348). At first it is a shallow groove just at the pole itself, but it soon spreads

over the pigmented hemisphere. The ends may progress at a uniform rate, or one may exceed the other in rapidity as in Petromyzon (Eycleshymer, loc. cit.).

At the first appearance of the furrow it is very shallow and perfectly smooth, and extends about 0.2 mm. on either side of the pole. It begins to deepen in two or three minutes, and at the same time minute wrinkles appear on either side (Plate I, figs. 1, 5, 7). If the eggs be placed upon a black surface in the sunlight these wrinkles are seen very distinctly as fine lines radiating from the pole (Plate II, fig. 31). The number and arrangement of these lines is not constant or definite either at this, or any subsequent period. The suggestion at once presents itself, that these wrinkles may be the foreshadowing of subsequent segmentations, and one egg was obtained in which the wrinkles seemed especially significant in their arrangement (Plate II, fig. 32). The entire lack of regularity in the size and arrangement of the wrinkles would, however, preclude any such idea, since the subsequent segmentations are very regular.

As the furrow gradually deepens and extends toward the yolk two changes are noticed :

1. New wrinkles appear along its sides ; these do not all radiate from the pole, but are inclined toward it at greater or smaller angles (Figs. 6, 8, 11).

2. The radial wrinkles first formed at the pole change considerably. Very fine and delicate at first they coalesce gradually into a few larger and deeper ones, which are sharply defined.

These fused ones may or may not occupy the position of one of the antecedent ones (Figs. 2, 3, 33, 34, 35). As the furrow progresses the number and position of the wrinkles changes constantly, new ones being formed and old ones disappearing. This is especially true of the finer ones ; some of the larger fused ones near the pole remain quite constant (Figs. 10–15).

This continual changing is best seen by making sketches of the wrinkles with a camera lucida at short intervals, as in the movements of the pseudopodia of Amœba (Figs. 1–4 and 10–15).

The whole appearance thus far is exactly as if the egg were covered by a very thin, but firm membrane, which was gradually pulled in toward the center at the groove. The remainder of the sphere being perfectly even, and with no chance for " give " at any point, in consequence of the uniform tension, the edges of the groove would necessarily be wrinkled, the wrinkles becoming more and more prominent as the groove deepened. The whole process takes place so slowly that the

most careful scrutiny fails to detect the actual motion, or to see evidences of any movement of the protoplasm within the egg, which might cause the wrinkling.

The furrow at the pole has become quite deep by the time its ends have reached the equator, in five or six minutes. The ends seem to stop here for a time, just at the border of the yolk area, while further changes take place in the pigmented portion. First the two edges of the groove approach each other at the pole, and seem to fuse slowly, the wrinkles entirely disappearing during the process. This fusion then extends in either direction along the groove for some distance, often half way to the equator, obliterating the wrinkles as it goes. By this means the groove may entirely disappear at the center while remaining near the periphery (Fig. 38).

At this point the furrow begins to enter the yolk area on either side, and at the same time the groove reappears along the center of the pigmented hemisphere. It now becomes very broad and deep. Indeed, it seems to reach clear through to the yolk, and its walls are considerably rounded on either side. But they are now smooth, so that the wrinkles remain in all fifteen or twenty minutes on the first furrow.

This first furrow divides the egg into two nearly equal parts (Figs. 3, 12, 28, 35). When its ends first reach the yolk area, where they stop for a time, as already noted, the two blastomeres thus formed are very much rounded at their ends, and diverge strongly from the groove. This is readily seen in the series given in figs. 1 to 4, but it becomes much more prominent after the reappearance of the groove—(Figs. 28 to 30). Under a higher magnifying power a wide space can now be seen at either end between the two segments. The floor of this space is triangular in shape and doubly curved, being concave from side to side, and convex antero-posteriorly. It is formed of light colored yolk, into which the pigment shades gradually around the borders. The two segments are thus rounded in a manner very similar to that of the first two blastomeres of a meroblastic egg.

As the furrow proceeds toward the inferior pole the space between the two pigmented segments diminishes, the borders of the furrow approach each other, and the surface of the egg becomes smooth once more, with the groove indicated merely by a narrow, faint line.

It remains in this condition some eight or ten minutes before the second cleavage begins, and this may be called its resting stage. The rapid closing of the groove previous to the appearance of the second furrow occurs also in Amblystoma (Eycleshymer).

The Second Cleavage.—The second furrows begin about forty or forty-five minutes after the first. They start *from the equator* and move toward the dark pole, and not vice versa as in Rana, Amblystoma, Petromyzon, and most of the amphibians. This was without exception in all cases noted (some thirty-five or forty). They form a right angle with the first groove, but do not always meet it at the same point (Figs. 19, 20). They are attended with the same appearance, fusion, and gradual elimination of wrinkles as the first furrow, with the slight difference that the wrinkles start at the equator instead of the pole, and are never quite as large. This is shown in figs. 16 to 18, in which both the wrinkles and the furrows can be seen approaching the pole.

The second furrows seem to reach the first over the dark hemisphere before they start toward the inferior pole. They start at about, but not necessarily exactly, the same time.

Here, too, there is the same rounding of the pigmented segments, though to a much less degree. This may be due to the fact that in this instance the furrows start from the periphery, and consequently the segments are more sharply defined there.

The Third Cleavage.—Half an hour after the appearance of the second set of furrows, and sometimes before they have reached the inferior pole, there are indications of the first horizontal cleavage. As seen from figs. 21–24 this occurs very much nearer the superior pole than the inferior. The furrows, accompanied as before by wrinkles along their edges, as seen in the figures, may start in each segment either from the first or second vertical, usually, however, from the second (Fig. 22).

They move much more rapidly than the preceding cleavages, and the entire process is completed in fifteen to eighteen minutes.

In all the eggs observed the grooves started usually at the second vertical in the two quadrants on the same side of the latter, and moved around toward each other at the first vertical.

They seldom met at the same point, but as a result of their formation the superior region was divided into four quite equal and nicely rounded cells, much smaller than the four inferior ones.

The only wrinkles formed upon the yolk area that could be detected by the most careful examination are the few which occur on the yolk side of this first horizontal groove. Schultze failed to detect any wrinkles whatever along this horizontal furrow, although he describes the other details with great exactness. This is all the more striking, because he both observed and figured them upon subsequent furrows up to the

32-cell stage. Other authors, with possibly a single exception, make no mention of them, except along the first furrow.

This confining of the wrinkles almost exclusively to the pigmented area is manifestly connected with the different organization of the two halves of the egg. The pigmented half is richer in protoplasm, and is to a higher degree under the influence of the cell nucleus; while the yolk has its protoplasm scattered about amongst the yolk granules, and is also further removed from the nucleus which lies in the pigmented half in the undivided egg (Hertwig). This results in the more rapid segmentation of the pigmented cells, and the presence of the wrinkles seems intimately associated with this rapidity of segmentation.

The Fourth Cleavage.—This appears from fifteen to twenty minutes after the third.

In this cleavage also the furrows started in every instance *from the periphery* of the four superior quadrants and move toward the pole, accompanied by wrinkles. These latter are now much smaller than heretofore, and are not easily detected under a low power. They are also very transitory, and disappear almost immediately. There is somewhat of a tendency in these furrows to run nearly parallel to the first or second vertical, recalling the conditions in teleosts (Figs. 26, 27).

There is a subsequent fusion and elimination of the furrows after each cleavage, as has already been noted in the first segmentation.

This elimination of the grooves, due to the fourth cleavage, leaves the pigmented pole of the egg divided into four cells of a totally different shape and arrangement from that of the four blastomeres resulting from the third cleavage (Fig. 37). After remaining thus for several minutes the furrows reappear, and the cells resume the shape seen in fig. 36.

From this point segmentation proceeds very rapidly, and in a manner exactly similar to that of other frog's eggs. The wrinkles have now become so small as to be seen only with the greatest difficulty and under a high power, and they disappear so quickly as to easily escape detection. But they are present at least up to the 128-cell stage, and appear, fuse, and disappear, as in the first cleavage.

Gastrulation begins about twelve hours after the first cleavage, and the blastopore closes at about the fifteenth hour. The neural folds appear and gradually fuse to form the neural canal as in Rana. By the end of the first day the embryo has elongated considerably, and the head is well differentiated.

The tail then becomes defined, the gill folds appear, and the eyes are seen as two minute black spots. And by the end of the third day the embryo escapes from the egg envelopes and swims about freely.

The yolk sac seems unusually large, and the tail is comparatively long at this period, but otherwise these tadpoles are externally like those of Rana and Hyla.

NATURE AND ORIGIN OF THE WRINKLES.

If the eggs be preserved during the first segmentation while the wrinkles are still present, and then sectioned parallel to a plane tangential to the superior pole, considerable additional light is thrown upon this process of wrinkling.

As has been noted in the eggs of other frogs the pigment is gathered into a thin surface layer over the superior hemisphere.

Houssay states that the pigment does not characterize this pole, but only happens to be there on account of the coincidence of its density with that of the surrounding protoplasm (Etudes d'Embryologie sur les Vertébrés, Archiv. de Zoöl. Exper., 1890). However this may be, during segmentation pigment appears along the sides of the furrows, so that eventually the resultant cells come to have a more or less definite pigment layer around their periphery, Houssay accounts for this by saying that there is an intimate relation between the activity of the cell and the presence of pigment. When the resting cell becomes active its granules become smaller, and pigment appears in them as the result of chemical action.

The presence of pigment, therefore, is the result of an increased activity in the cell.

But Bambeke tells us that "the cortical layer, when it enters the interior of the protoplasm, is not entirely employed in limiting the spheres of new formation. In fact, I find masses of pigment whose presence can only be explained by considering them as debris from the cortical layer, which has penetrated into the protoplasm" (Fractionnement de l'Oeuf des Batraciens, Archiv. de Biologie, Vol. I, p. 346, footnote).

An examination of sections of these Chorophilus eggs shows a similar occurrence.

In addition to the pigment layer which borders the first segmentation furrow, and which is somewhat thicker near the centre of the section, there is also a lunate mass extending downward vertically from the superior pole on either side of the furrow (Fig. 39) and in immediate contact with it. This mass can be traced in the sections from the surface layer, in which it has very little area, down somewhat beyond the bottom of the furrow, where it spreads out laterally and is lost in the surrounding protoplasm. In this particular egg the mass does not

extend quite to the level of the nuclei, the position of the latter in the figure having been taken from subsequent sections in the series. This mass, however, is not to be regarded as "debris," but its presence is due to a definite cause to be explained later. The remainder of the section is occupied by homogeneous protoplasm filled with rather small yolk granules, and surrounded by the thin, transparent vitelline membrane.

Under a high power (Fig. 40) the wrinkles appear as deep sinuses extending obliquely into the protoplasm, and bordered by a thick layer of pigment. These sinuses are angular and irregular in outline, and often cantract at their inner ends into long, narrow slits, with rather distinct walls. We can now see what it was impossible to detect from a surface view, namely, that the wrinkles are compound.

The larger, principal ones have secondary, smaller ones extending outward from their sides, approximately at right angles. Schultze observed and figured these compound wrinkles in his surface views of R. temporaria, and adds another detail which I have been unable to find in the Chorophilus eggs, viz., the breaking-up of a single wrinkle at its peripheral end into several smaller ones arranged radially from a common point.

The pigment usually fills the projecting protoplasm between adjacent sinuses. It is also much thicker in the region of the wrinkles than elsewhere along the furrow, as can be seen in fig. 40.

In view of these different facts, therefore, it seems evident that there is an intimate relation between the wrinkles and the pigment—and that both may be results of the same cause. It remains to ascertain what this cause is, if possible.

According to Schultze the egg is a single cell, and just as cellular division is brought about by the contractility of protoplasm, so is the segmentation of the egg due to the same cause. These contractions originate at the point where the furrow begins, and are at first confined to a very small area. Since the cortical portion of the egg protoplasm possesses a glutinous consistency, it is not to be wondered at that folds or wrinkles appear at the same time with the furrow, in its immediate vicinity, and at right angles to it. These subsequently disappear in consequence of the difference in contractility between the outer and inner protoplasm, due to their different consistency.

It is exceedingly difficult to understand how compound wrinkles, of such a nature as we have just described, could be produced by the simple contraction of a viscous cortical layer of protoplasm, especially if that contraction *starts from a fixed* point in the layer. Indeed, how

could it produce any wrinkles at all in the layer itself? Would it not rather tend to flatten the superior pole and stretch the viscous layer tightly over the underlying protoplasm in a manner similar to the action of the muscles of the diaphragm during respiration?

As to the difference in contractility between the outer and inner protoplasm, it is evident that if this is to smooth out the wrinkles, the difference must be in favor of the outside layer, and also the contraction must be *at right angles to the length of the wrinkles.* That means in the present instance that it must be parallel to the first furrow. Neither of these conditions seem possible, and we thus find both explanations inadequate when confronted by the facts in the case. They both fail to account for the mass of pigment under the superior pole also.

Bambeke states that "the entrance of cortical pigmented masses into the interior of the egg . . . can only be explained by admitting the existence of contractions in the ovular protoplasm during segmentation."

According to the interpretation of the present day, cell division is brought about by means of some force or forces acting along the length of the segmentation spindle. In the present instance this spindle was formed between the two nuclei represented in fig. 39, which lie some distance below the surface of the egg.

If we interpret Bambeke's "contraction of the ovular protoplasm" to be identical with this working force of the segmentation spindle, it will explain the presence of the lunate mass of pigment directly over the spindle, and will help us to understand the presence of a pigmented layer on either side of the segmentation furrows. But it does not explain in any way the formation of the wrinkles.

Reichert's wonderfully inconsistent explanation of the origin of the wrinkles is quoted, and sufficiently commented upon by Schultze, in the paper already referred to.

In view of the fact, therefore, that we have no explanation which can stand the test of our present knowledge of cell division, we venture to offer the following:

We agree with Schultze that the external pigmented layer is necessarily somewhat denser that the internal protoplasm.

Modern research indicates that this layer is drawn inward in some way, by the forces working along the segmentation spindle, to form the furrow which lies over the equator of the spindle. The bottom of the furrow, thus formed by an infolding of the surface layer, describes an arc which becomes shorter and shorter as the furrow deepens.

This shortening of the arc must result in one of two things. The bottom of the furrow may remain of the same length as at first, and

make up for the shortening of the arc by protruding a little from the surface of the egg at either end of the furrow. Such a condition is admirably shown when one creases the top of a soft felt hat, and would necessarily be even more manifest if the hat were filled with a viscous fluid.

An examination of any of the surface views will show that this is not the case with these Chorophilus eggs, but that the first indication of the groove at either extremity is a slight hollowing in of the surface, and not a bulging outward. This view is confirmed by a study of the sections.

The other alternative is that the shortening of the arc must result in longitudinal condensation along the bottom of the furrow, starting at the center and increasing as the furrow progresses.

Such a shortening or contraction along the bottom of the furrow would very naturally throw its sides into folds or wrinkles at right angles to its length. The pigment layer in contact with these walls would also be thickened in the region of the wrinkles, and toward the center of the groove.

Since the condensation starts at the center and advances in both directions with the progress of the furrow, the wrinkles would be arranged somewhat radially about the superior pole. This progressive contraction also accounts for the successive appearance and disappearance of wrinkles, and for the confluence of smaller into larger ones.

As the sides of the furrow begin to fuse into the permanent segmentation plane or cell-wall the wrinkles disappear through the gradual readjustment of former relations.

Indeed, the whole phenomenon seems very largely dependent on the rapidity of segmentation and the consequent sudden disturbance of normal relations before the different portions can adjust themselves to their new conditions.

This fact will serve to explain why the wrinkles show so prominently in this particular species, which has a very rapid development, and also why the conditions under which they were examined—the transference from ice to tepid water and the placing of the eggs in strong sunlight—were especially favorable.

It may also suggest a reason why one observer has failed to detect wrinkles in the eggs of a given species, while another, working under more favorable conditions, has seen and described them. And it will in a measure account for the absence of wrinkles on the yolk hemisphere, since segmentation is very much slower there.

If our explanation is a correct one there ought to be no wrinkles at all along the bottom of the groove, while they should be present and have their greatest depth about half way between the bottom and the surface.

The sections show that this actually occurs. Fig. 39 is a section cut just at the level of the bottom of the groove, and shows no trace of any wrinkles, nor are there any in the two or three preceding sections.

They then appear and gradually increase in size up to the level of fig. 40, which is a magnified portion of the same groove about half way to the surface.

The problem of the compound nature of the wrinkles finds its solution in the fact that there must be a condensation along the bottom of the larger wrinkles, in all respects similar to that in the groove, and due to the same cause, though, of course, on a very much smaller scale.

But in this instance the condensation would proceed in only one direction, and hence we find the secondary wrinkles all inclined in the same direction to the principal ones, just as we have already observed, and as Schultze has so finely figured.

Summary.—1. Subjection to a temperature of 0° C. for a period of eight hours completely arrests all development for the time being, but results, on the subsequent restoration of ordinary conditions, in a cleavage more rapid than that of normal eggs.

2. Segmentation, at least up to the 128-cell stage, is accompanied by the formation, fusion and subsequent elimination of well defined wrinkles along the sides of the furrows in the pigmented area. There are no wrinkles on the yolk, except along the inferior border of the third cleavage furrow.

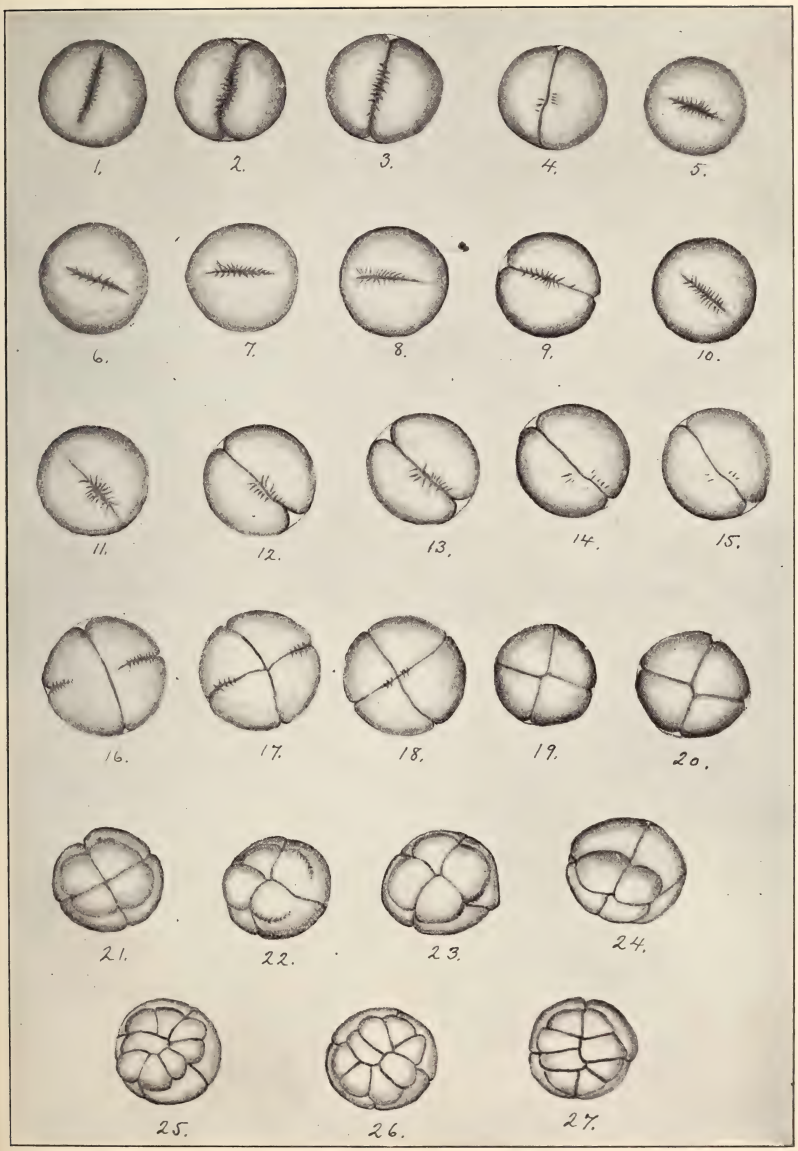
3. As seen in an examination of cross-sections these wrinkles are compound in nature, the larger, principal ones having smaller secondary ones along their sides.

4. The wrinkles on the first furrow are arranged somewhat radially about the superior pole. On subsequent furrows they are inclined at an angle toward the point where the furrow starts.

5. The pigment which borders the segmentation furrows forms a thicker layer in the region of the wrinkles than elsewhere along the groove, thus showing an intimate relation between the two.

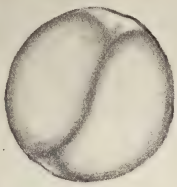
6. The probable cause of the wrinkling is to be found in the condensation along the bottom of the groove, which results from the shortening of the arc, and is a necessary consequence of the infolding of the surface layer to form the groove.

PLATE XIII.



Segmentation of Rana.

PLATE XIV.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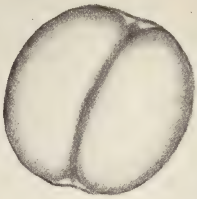
29.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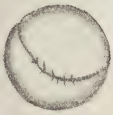
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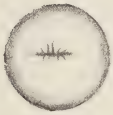
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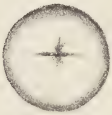
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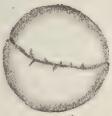
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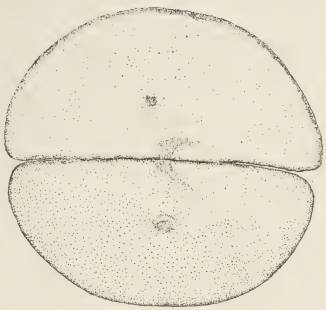
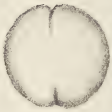

39


37.


36.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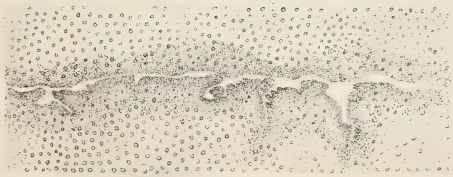
40

Segmentation of Rana.

7. The second and fourth grooves *start from the periphery* and move toward the pole.

8. The blastomeres are more rounded and the segmentation furrows are deeper than those in most frog's eggs.

9. The development is very rapid; gastrulation begins within twelve hours, and the tadpole escapes from the egg during the second or third day.—Charles B. Wilson.

Explanation of Plate I.

All figures drawn with Zeiss Camera x 16 diam.

Figs. 1–4. Successive stages in cleavage of living egg at intervals of 3 mins., 3 mins., 2 mins.
Figs. 5, 6. Stages in cleavage of second living egg.
Figs. 7–9. Stages in cleavage of third living egg—intervals, 2 mins., 4 mins.
Figs. 10–15. Stages in cleavage of fourth living egg—intervals, 2, 6, 3, 4, and 5 mins.
Figs. 16–18. Stages in second cleavage of third egg.
Figs. 19, 20. Variations in second cleavage.
Fig. 21. Eight-cell stage of first egg.
Figs. 22–24. Stages in third cleavage of second egg.
Figs. 25–27. Variations in sixteen-cell stage.

Explanation of Plate II.

All figures drawn with Zeiss Camera.

Figs. 28–30. First cleavage under higher magnification to show rounded blastomeres.
Figs. 31, 32. Beginning of first cleavage, showing radiating wrinkles at pigmented pole.
Figs. 33–35. Variations in wrinkles on the first furrow.
Fig. 36. View of an egg during the fourth cleavage.
Fig. 37. The same egg four minutes later.
Fig. 38. Fusion and partial disappearance of the first groove.
Fig. 39. Horizontal section of an egg during the first segmentation, taken at the level of the bottom of the furrow. Nuclei added from the fourth section below this.
Fig. 40. A portion of the same furrow about half way between its bottom and the surface of the egg, more highly magnified.

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